

10/656, 805

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Serial No. : 10/656,805
Filed : September 5, 2003
Page : 4 of 28

Attorney's Docket No.: 10448-215010 / MPI98-
129CN1A

and XhoI sites of pcDNA3 (Invitrogen, San Diego, CA) to create the mammalian expression plasmid pCD5MCPRB. The CD5-CCR2b fragment was subcloned into the BamH I-Not I site of pCDEF3 (Goldman *et al.*, (1996) *Biotechniques* 21:1013-1015), and this construct was designated CCR2bDEF3. In this expression vector, the expression of the inserted gene is driven by the EF-1 α promoter.

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4/13/09

Please replace the paragraph beginning at page 49, line ¹⁰9 with the following amended paragraph:

For the staining of cultured transfectant cell lines 0.5×10^6 cells in 50 μ l were resuspended in PBS + 1% FCS in a 96 well polystyrene V-bottom plate. 50 μ l of primary antibody supernatants or HT medium (negative control) were added, and the samples were incubated at 4°C for 30 min. 100 μ l of PBS were added and the cells were pelleted by centrifugation and washed once with PBS. The pellet was resuspended in 100 μ l PBS + 1% FCS containing FITC-conjugated goat anti-mouse IgG antibody (a 1:250 dilution) and incubated for thirty minutes at 4°C in the dark. The cells were washed twice with PBS, resuspended in PBS, and analyzed by flow cytometry cytometry with a FacScan FACScan® cytometer using the CellQuest™ software (Becton-Dickenson Dickinson). Cells were fixed with PBS/1% formaldehyde if they were not to be analyzed the same day. Monoclonal antibodies 1D9 and 8G2 stain CCR2 transfectants but not CCR1 or CCR5 transfectants (FIGS. 1A-1O).

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4/13/09

Please replace the paragraph beginning at page 49, line ²³9 with the following amended paragraph:

100 μ l whole blood was mixed with 100 μ l of 1D9 antibody hybridoma supernatants or HT medium (negative control) and incubated at 4°C for 30 min. After one wash with PBS, 100 μ l FITC-conjugated goat anti-mouse IgG antibody (a 1:250 dilution) was added to each sample and incubated for 30 min. at 4°C in the dark. Samples were then washed once with PBS if a second color staining is to be done, otherwise washed twice more in PBS. For two color staining 5 μ l of mouse serum was added to the cell pellets after the single wash, mixed, and